● PRINTER RUSH ●
(PTO ASSISTANCE)

Application : 09/661848 **Examiner :** Ketter **GAU :** 1636

From: T. McGill **Location:** (IDC) FMF FDC **Date:** 4-26-05

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[RUSH] **MESSAGE:** Specification has a missing SEQ ID No. on page 52, line 20.

Thankyou

[XRUSH] **RESPONSE:** corrected

INITIALS: KP

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REV 10/04

substrate adhesion properties or other morphological changes that can be readily detected by a person having ordinary skill in the art, for example by using light microscopy. As another example, cells undergoing apoptosis may exhibit fragmentation and disintegration of chromosomes, which may be apparent by microscopy and/or through the use of DNA-specific or chromatin-specific dyes that are known in the art, including fluorescent dyes. Such cells may also exhibit altered plasma membrane permeability properties as may be readily detected through the use of vital dyes (*e.g.*, propidium iodide, trypan blue) or by the detection of lactate dehydrogenase leakage into the extracellular milieu. These and other means for detecting apoptotic cells by morphologic criteria, altered plasma membrane permeability and related changes will be apparent to those familiar with the art.

In another embodiment of the subject invention method wherein the indicator of altered mitochondrial function is a cellular response to an apoptogen, cells in a biological sample may be assayed for translocation of cell membrane phosphatidylserine (PS) from the inner to the outer leaflet of the plasma membrane, which may be detected, for example, by measuring outer leaflet binding by the PS-specific protein annexin. (Martin et al., *J. Exp. Med. 182*:1545, 1995; Fadok et al., *J. Immunol. 148*:2207, 1992.) In still another embodiment of this aspect of the invention, a cellular response to an apoptogen is determined by an assay for induction of specific protease activity in any member of a family of apoptosis-activated proteases known as the caspases (see, *e.g.*, Green et al., 1998 *Science* 281:1309). Those having ordinary skill in the art will be readily familiar with methods for determining caspase activity, for example by determination of caspase-mediated cleavage of specifically recognized protein substrates. These substrates may include, for example, poly-(ADP-ribose) polymerase (PARP) or other naturally occurring or synthetic peptides and proteins cleaved by caspases that are known in the art (*see, e.g.*, Ellerby et al., 1997 *J. Neurosci. 17*:6165). The synthetic peptide Z-Tyr-Val-Ala-Asp-AFC ~~(SEQ ID NO:__)~~, wherein "Z" indicates a benzoyl carbonyl moiety and AFC indicates 7-amino-4-trifluoromethylcoumarin (Kluck et al., 1997 *Science 275*:1132; Nicholson et al., 1995 *Nature 376*:37), is one such substrate. Other non-limiting examples of substrates

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